REMARKS:

The specification has been amended to add the language "in which *in vivo* biological activity was determined by the polycythemic mouse bioassay." This amendment is supported by the application as originally filed at page 29, paragraph [0091] of the present application as originally filed. In this paragraph, Satake et al. 1990, *Biochim. Biophys. Acta* 1038:125-9 is incorporated in its entirety for its description of molecular biological techniques for generating erythropoietin derivatives and testing them for *in vivo* biological activity on erythrocytes (*i.e.*, erythropoietic activity) using the polycythemic mouse bioassay (see Satake, page 126, col. 2: "in vivo biological activity was determined by the exhypoxic polycythemic mouse bioassay."

Claims 1-5, 8-46, and 53-54 were pending in the instant application. Claims 1-5 were withdrawn from consideration. Claims 8-46 and 53-54 were under examination.

Claims 8 has been amended to incorporated the limitations of claim 11 except that several of the modifications formerly recited in claim 11 were not introduced in claim 8. Claim 8 has also been amended to recite "inflammatory disease." Support for this amendment can be found in the application as originally filed at paragraph [0139]. Claim 8 has further been amended to specify tests for determining *in vivo* erythropoietic and tissue protective activity. In particular, the modified erythropoietins have: (a) reduced *in vivo* erythropoietic activity compared to native erythropoietin as determined by the exhypoxic polycythemic mouse bioassay; and (b) tissue protective activity *in vivo* as determined by the middle cerebral artery occlusion test. Support for this amendment is found at paragraph [0091] of the specification as originally filed, and at paragraphs [0060] and [0061]; paragraph [0235]; and drawing sheets 7 and 8.

Claims 12, 13, 24, 26-28, 30, 31, 33, 36, 38, 40, 41, 43, 45, 53, and 54, all of which depend from claim 8, have been revised to reflect the amendment to claim 8.

Claims 11 and 17-23 have been canceled without prejudice. Applicants reserve the right to prosecute the subject matter of the canceled claims in one or more related continuation, continuation-in-part, or divisional applications.

New claim 55 has been added. Support for new claim 55 can be found in the specificaiton as originally filed at p. 54, *l.* 12.

No new matter has been introduced. Claims 1-5, 8-10, 12-16, 24-46, and 53-55 will be pending upon entry of the present remarks.

Attorney Docket Number

Applicants request that the U.S. Patent and Trademark Office revised its records to reflect the new attorney docket number of the present application: "10165-037-999." A Revocation and Power of Attorney bearing the new attorney docket number was submitted on September 28, 2007.

SUMMARY OF THE SUBSTANCE OF THE INTERVIEW

Applicants thank Examiner Cherie Michelle Woodward for the courtesy extended during the interview conducted on May 21, 2008 via video conference ("the Interview"). Also present at the Interview were Drs. Anthony Cerami and Michael Brines, two of the inventors of the instant application, Frederick J. Hamble, Esq., of Warren Pharmaceuticals, Inc., and applicants' representatives Laura A. Coruzzi, Eileen E. Falvey, Markus Bergauer, and Sebastian Martinek of Jones Day.

During the Interview, the rejections under 35 U.S.C. § 112, first paragraph, made in the Office Action dated December 27, 2007 (the "Office Action") were discussed. At the Interview, Applicants presented results of experiments to support enablement commensurate with the full breadth of the claims.

Dr. Cerami explained that EPO has two biological activities—the promotion of blood formation and the protection of tissue. The erythropoietic activity and the tissue-protective activity of EPO can be separated from each other because these two activities are mediated through different receptors. The elimination of EPO's erythropoietic activity through chemical modification of EPO results in improved tissue-protective forms of EPO. In particular, clinical use of these improved forms of EPO eliminates the risk of thrombosis associated with the use of unmodified EPO.

Dr. Cerami further discussed that tissue-protective forms of EPO also suppress inflammation. EPO has been shown to act synergistically with Methylprednisolone, an anti-inflammatory agent, in an animal model for multiple sclerosis, experimental autoimmune encephalomyelitis (EAE).

Dr. Coruzzi stated that Applicants propose to amend the claims to recite (i) certain chemical modifications of EPO; and (ii) functional assays for tissue-protective and erythropoietic activity of the chemically modified EPO.

The Examiner indicated that the evidence, arguments, and any amendments would be considered once presented in response to the outstanding Office Action. The present amendment, the evidence provided in a declaration by Michael L. Brines, M.D., Ph.D. dated October 5, 2007 ("Brines I") and a second declaration by Dr. Brines dated February 20, 2008 ("Brines II"), and the remarks herein reflect the discussion during the Interview.

THE CLAIM REJECTIONS UNDER 35 U.S.C. § 112, FIRST PARAGRAPH, ENABLEMENT, SHOULD BE WITHDRAWN

Claims 8-46, 53, and 54 have been rejected under 35 U.S.C. § 112, first paragraph, as failing to comply with the enablement requirement. Applicants assert that the specification is enabling for the claimed invention and that the indicated claims should be allowed. For ease of reference, Applicants have organized their response in two parts as the Office Action dated December 27, 2007 is structured.

Part I

Claims 8-10, 13-16, 43-46, 53, and 54 were rejected for lack of enablement because the claims read on any generic tissue protective cytokine. Without agreeing to the Examiner's rejection and solely to expedite the prosecution of this application, Applicants have amended claim 8 to incorporate the limitations of claim 11. Instead of "a tissue protective cytokine," amended claim 8 recites an erythropoietin with specific modifications. Accordingly, Part I of the enablement rejection is moot in view of claim 8. Dependent claims 9-10, 13-16, 43-46, 53, and 54 all ultimately depend from claim 8 and incorporate the limitations of claim 8 via their dependencies. Accordingly, Part I of the enablement rejection is also moot with respect to these claims.

Part II

Claims 11-12, 17-42, 53, and 54 were rejected for lack of enablement.

Sufficient Guidance For EPO Modifications

The Examiner states that the specification fails to provide sufficient guidance with respect to the modifications of EPO such that EPO retains its function.

First, it is noted that claim 8 has been amended to recite erythropoietin with specific chemical modifications in place of the term "tissue protective cytokine." In addition, several modifications of EPO's glycosylation pattern and truncation of EPO, are no longer recited in amended claim 8. Thus, the rejections that were based on these claim terms are moot.

The amended claims identify the chemically modified erythropoietin ("EPO") by its structure—the amino acid sequence of EPO was well know and specific chemical modifications are listed in the claim. The amended claims further identify the chemically modified EPO by its function—(a) a reduced level of *in vivo* erythropoietic activity compared to native erythropoietin as determined by the exhypoxic polycythemic mouse bioassay,[1] and (b) tissue protective activity *in vivo* as determined by the middle cerebral artery occlusion test.[2] In view of the guidance provided in the present application and the common general knowledge in the art, any experimentation that may be required to identify suitable chemically modified forms of EPO is merely routine. Applicants submit herewith a declaration by Michael L. Brines, M.D., Ph.D. dated October 5, 2007 ("Brines I") and a second declaration by Dr. Brines dated February 20, 2008 ("Brines II") as evidence that no undue experimentation would be required to practice the claimed methods. Brines I and Brines II were previously submitted in related U.S. Application No. 10/185,841 ("the '841 Application"). The present application and the '841 application claim benefit of priority of PCT/US01/49479 filed December 28, 2001. Brines I refers to support in the '841 Application by page and line numbers. The chart set forth in Exhibit E provides the corresponding page and line numbers in the present application. In addition, the first reference cited in ¶ 26 of Brines I is now published. The full cite is Imamura, 2007, Biochem. Biophys. Res. Comm. 353:786-792.

For the Examiner's convenience, Applicants have structured the discussion regarding enablement as follows: (1) Chemical Modification of EPO; (2) Non-Erythropoietic Forms of EPO; (3) Tissue-Protective Forms of EPO; (4) EPO Activity in Various Tissues; and (5) Protection and Regeneration of Tissue, Prevention of Injury, and Restoration of Function.

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[1] For ease of reference, forms of EPO with reduced *in vivo* erythropoietic activity will be referred to in this response as "non-erythropoietic" forms of EPO.

[2] For ease of reference, forms of EPO with tissue protective activity *in vivo* as determined by the middle cerebral artery occlusion test will be referred to in this response as "tissue-protective" forms of EPO.

(1) Chemical Modification of EPO

EPO's amino acid sequence was well-known in the art (Brines I, ¶6). Protein modifications such as the types of modifications recited in claims 35 and 37 were disclosed in the present application and it was merely routine to carry out these methods for modifying proteins (Brines II, ¶¶7-11). Thus, no undue experimentation would be required to generate the chemically modified forms of EPO as recited in claims 35 and 37.

(2) Non-Erythropoietic Forms of EPO

The generation of non-erythropoietic forms of EPO does not involve undue experimentation because (i) EPO can be chemically modified as discussed in Section (1) above and the erythropoietic activity can be tested using a routine assay--the exhypoxic polycythemic mouse bioassay; (ii) guidance in the specification; and (iii) guidance in the art.

(i) Routine Assays

The exhypoxic polycythemic mouse bioassay is one of various assays that have been routinely used to test erythropoietic activity of candidate compounds (Brines I, ¶14). Thus, merely routine assays would be used to verify the reduced level of *in vivo* erythropoietic activity of a chemically modified EPO.

(ii) Guidance in the Present Application

The present application identifies at p. 29, ¶ [0091], four regions in the EPO protein (SEQ ID NOs:1-4) that can be modified to reduce EPO's erythropoietic activity. In particular, it is disclosed that basic amino acid residues, arginine and lysine, are modified. In addition, the application teaches that areas surrounding SEQ ID NOs:1-4 can be modified to achieve reduced erythropoietic activity (¶ [0091]).

(iii) Guidance in the Art

Modifications that result in reduction of EPO's erythropoietic activity were known in the art (Brines I, ¶15). Further, guidance with respect to what kinds of modifications would reduce EPO's erythropoietic activity are set forth, *e.g.*, in Satake, 1990, Biochem. Biophys. Acta 1038:125-129 ("Satake," cited at p. 29, ¶ [0094], of the present application). In particular, Satake reported that modification of lysine residues to neutral or negative charges results in non-erythropoietic EPO (Brines I, ¶15).

(3) Tissue-Protective Forms of EPO

As discussed during the interview of May 21, 2008, EPO's erythropoietic activity can be reduced without affecting its tissue-protective or anti-inflammatory activity because these two separate activities are mediated through different receptors (Brines II, ¶6-7). Thus, chemical modification of, *e.g.*, those regions of the EPO protein that are responsible for binding to the receptor that mediates erythropoiesis will reduce EPO's erythropoietic activity while the modified EPO remains fully tissue-protective.

Applicants demonstrate for a wide variety of different chemically modified EPOs that tissue-protective activity is retained. These examples include: asialoEPO (¶234); phenylglyoxalEPO (¶234); iodo-EPO (¶250); biotinylated EPO (¶253); carbamylated asialoEPO (¶272); succinylated asialoEPO (¶272); acetylated asialoEPO (¶272); iodinated asialoEPO (¶272); carboxymethylated asialoEPO (¶272); carbamylated EPO (¶272); acetylated EPO (¶272); and N^{ε}-carboxy methyl EPO (¶272).

In addition, the middle cerebral artery occlusion test is a routine assay that can be used to verify that a chemically modified EPO has tissue-protective activity. No undue experimentation would be required to prepare chemically modified EPOs with reduced erythropoietic activity and to subject these EPOs to the middle cerebral artery occlusion test to verify that these forms of EPO have retained their tissue-protective activity.

The Office Action dated December 27, 2007 alleges that the present application is silent regarding the biological activity of EPO having at least one modified lysine residue. See, the sentence spanning pages 11 and 12 of the Office Action dated December 27, 2007. Applicants disagree. Carbamylation of EPO, a form of lysine modification, is taught in the specification as originally filed at ¶[00237]. The tissue-protective activity is retained in carbamylated EPO. See, ¶[00272]. That tissue-protective forms of EPO have anti-inflammatory effects has also been shown in the present specification. See Example 12, beginning at p. 110.

(4) EPO Activity in Various Tissues

Brines II, and references cited therein, provide ample evidence that the claimed methods can be used in a wide variety of mammalian cells, tissues and organs. As discussed in ¶¶ 6-8 of Brines II, the receptor that mediates EPO's tissue protective activity is expressed

in many tissues such that EPO can exert its tissue protective, and anti-inflammatory, function in these various tissues.

For example, as described in ¶¶ 10-15 of Brines II, chemically modified EPOs have been shown to provide tissue protective effects in numerous cell types including brain, spinal cord, peripheral nervous tissue, heart, kidney and skin.

In addition, EPO has been shown to provide tissue protective activity in numerous other types of cells, tissues and organs, including cochlea, striated muscle, endothelium, hair follicles, bone, intestine, lung, and liver. Brines II, ¶¶ 16-23. As explained in ¶¶ 6 and 7 of Brines II, chemically modified, tissue protective forms of EPO are effective in any tissue in which EPO is tissue protective, because such chemically modified forms of EPO are deficient only in the erythropoietic function of the EPO molecule (*e.g.*, by interfering with the EPO's ability to interact with the classical EPO receptor), and are not deficient in the tissue protective function of the molecule, which requires its interaction with a different receptor, a heteromer of the classical EPO receptor and the common beta receptor (the "Tissue-Protective Receptor Complex"). Brines II, ¶¶ 5-7. Thus, by this reasoning, chemically modified, tissue protective EPO molecules can provide tissue protective activity and anti-inflammatory activity in numerous other types of cells, tissues and organs, including cochlea, striated muscle, endothelium, hair follicles, bone, intestine, lung, and liver.

Moreover, chemically modified, tissue protective forms of EPO would be effective in any cell, tissue, or organ that expresses the Tissue-Protective Receptor Complex, which "may be found in all tissues." Brines II, ¶ 8. Tissue types that are known to express the Tissue-Protective Receptor Complex include, but are not limited to, endothelial cells, myocardiocytes, macrophages, retinal cells, cells of the adrenal cortex and medulla, small bowel, spleen, liver, kidney and lung, as well as cells of the central nervous system, such as neurons and glial cells, and astrocytes. Brines II, ¶ 8, and Appendix B.

The universal nature of EPO-mediated suppression of inflammation is also demonstrated by EPO's ability to prevent TNF production (see, ¶323 of the present application).

The Relevance of Cuzzoncrea and Savino

Applicants had submitted a copy of Cuzzocrea *et al.*, 2005, Arthritis & Rheumatism 52:940-950 ("Cuzzocrea;" Exhibit F) with their response of September 28, 2007. Cuzzocrea

provides evidence that the anti-inflammatory effect of EPO is not limited to the EAE model but rather is a general property of tissue-protective forms of EPO. This is consistent with the teaching in the present application that EPO suppresses the production of the pro-inflammatory cytokine TNF.

Applicants also submitted a copy of Savino *et al.*, 2006, Journal of Neuroimmunology 172:27-37 (Exhibit D; "Savino") with their response of September 28, 2007 to demonstrate that chemically modified forms of EPO that are tissue-protective also retain their anti-inflammatory effect.

Diem Demonstrates Synergism

Applicants submitted a copy of Diem *et al.*, 2005, Brain 128:375-385 (Exhibit C; "Diem") as evidence that EPO and an anti-inflammatory agent act synergistically (see, *e.g.*, p. 376 of Diem, last paragraph of the Introduction).

The synergistic effect is particularly compelling in the results presented in Figure 1F of Diem. Only the combined therapy of EPO and MPred restores visually evoked potentials in the visual cortex. Neither EPO alone or MPred alone restores visually evoked potentials. Thus, Diem demonstrates the synergistic effect of EPO together with MPred.

Gorio Is Consistent With The Workability Of The Claimed Methods

The Examiner cites Gorio *et al.*, 2005, PNAS 102:16379-16384 ("Gorio") as evidence for the contention that the claimed method is allegedly not workable. Gorio, however, is inapplicable because Gorio relates to spinal cord injury and not to inflammatory disease as presently claimed. In addition, the anti-inflammatory agent used by Gorio by itself also failed to exhibit a beneficial effect in Gorio's model system. In Gorio, methylprednisolone ("MPSS") was used alone and in combination with erythropoietin. Gorio shows that MPSS alone did not have a significant effect on the spinal cord injury model used in Gorio. Table 1, at p. 16381 of Gorio shows the percentage of spared tissue after a spinal cord injury. Row 2 of the table shows that EPO alone has a protective effect, *i.e.*, the percentage of spared tissue is increased. However, MPSS alone did not increase the percentage of spared tissue, *i.e.*, MPSS alone did not improve the outcome after the injury. Figure 7A, at p. 16383 of Gorio, shows the recovery of hindlimb function after the injury. MPSS alone had no beneficial

effect on hindlimb function. Thus, on an anatomical level (spared tissue) and a behavioral level (locomotor activity), MPSS did not ameliorate a symptom associated with the inflammation.

THE CLAIM REJECTIONS UNDER 35 U.S.C. § 112, FIRST PARAGRAPH, WRITTEN DESCRIPTION, SHOULD BE WITHDRAWN

Claims 8-10, 13-16, 43-46, and 54 have been rejected under 35 U.S.C. § 112, first paragraph, as failing to comply with the written description requirement because the claims read on any generic tissue protective cytokine. Amended claims 8-10, 13-16, 43-46, and 54 satisfy the written description requirement as discussed below. Without agreeing to the Examiner's rejection and solely to expedite the prosecution of this application, Applicants have amended claim 8 to incorporate the limitations of claim 11. Instead of "a tissue protective cytokine," amended claim 8 recites an erythropoietin with specific modifications. Accordingly, the written description rejection is moot in view of claim 8. Dependent claims 9-10, 13-16, 43-46, 53, and 54 all ultimately depend from claim 8 and incorporate the limitations of claim 8 via their dependencies.

Accordingly, the written description rejection is also moot with respect to these claims, and Applicants request that the rejection of claims 8-10, 13-16, 43-46, and 54 under 35 U.S.C. § 112, first paragraph, be withdrawn.

CONCLUSIONS:

Applicants respectfully request that the foregoing remarks and amendments be made of record in the file history of the instant application. Applicants estimate that the remarks and amendments made herein place the pending claims in condition for allowance.

by: Sebastian Hartwick
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Respectfully submitted,

Date: June 27, 2008

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